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EVALUATION OF THE ECOLOGICAL STATUS OF A STREAM IN MIDDLE TENNESSEE USING PERIPHYTON CHARACTERISTICS

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ABSTRACT

The upper reach of Big McAdoo Creek in the Lower Cumberland River Watershed in Middle Tennessee is listed as unimpaired by nutrient enrichment by the Tennessee Department of Environment and Conservation and the United States Environmental Protection Agency. The primary objective of this research was to determine if the reach should remain listed as unimpaired. Water quality was evaluated by examining concentration of soluble reactive phosphorous, characteristics of periphyton, and the composition and structure of the diatom assemblage. An oligotrophic concentration of soluble reactive phosphorous of the water and mesotrophic concentrations of photoautotrophic periphyton indicate the stream site sampled was not impacted by nutrient enrichment. Oligotrophic to mesotrophic values for the Siltation Index, Organic Pollution Index, and Pollution Tolerance Index for the diatom assemblage indicate the assemblage was not negatively impacted by siltation, high concentrations of dissolved organics, or nutrient enrichment, respectively. The results indicate the upper reach of Big McAdoo Creek should have its water-quality status remain as unimpaired.

Knowledge of the effects of nutrient concentration on the composition and structure of photoautotrophic periphyton is essential to understand the impact of eutrophication on shallow lotic systems. Nutrient enrichment and sediments from nonpoint sources are most responsible for the biological impairment of United States waters (Irvine & Murphy 2009). Assessments of nutrient and sediment pollution are prerequisites to developing watershed management plans to protect aquatic ecosystems (Smucker & Vis 2009). Biological monitoring is essential to characterize and quantify the influences of water quality. Periodic water sampling for chemical analyses alone may not reveal the impact of nonpoint-source pollution because pollutants from nonpoint sources often enter waterways in pulses. In addition, chemical analyses do not reveal the impacts of pollution on biological integrity (Taylor et al. 2007).

Photoautotrophic periphyton are the most important primary producers in the majority of wadeable streams (Lambert & Steinman 1997). Nutrient enrichment of streams changes photoautotrophic periphyton characteristics and affects whole-stream ecological relationships. The composition, biomass, and physiological status of photoautotrophic periphyton are excellent indicators of water quality and are used universally to follow changes in aquatic environments (Eaton et al. 2005). High concentrations of chlorophyll *a* as a measurement of the biomass of photoautotrophic periphyton is a hallmark of eutrophication (Khan & Ansari 2005). Because chlorophyll-*a* concentration is influenced by many abiotic and biotic characters of a stream reach, measurements of chlorophyll-*a* concentration alone may not be adequate to demonstrate impairment by nutrient enrichment (Kurle & Cardinale 2011).

Diatoms are the focus of most studies characterizing impacts of eutrophication on periphyton composition because more autecological information exists for diatoms relative to other algae (Smucker & Vis 2013). Information from the composition of diatoms can be used to support proposed restoration and conservation policies because diatom assemblages in impaired water often reflect the nature of impairment (Smucker & Vis 2009). Evaluation of diatom composition is a standardized protocol for monitoring changes of water quality in many European countries and states including Oklahoma, Montana, Kentucky, and Texas (Stevenson et al. 2008).

Big McAdoo Creek is a major tributary of the Lower Cumberland River Watershed and joins the Cumberland River approximately 20 km south of Clarksville, Tennessee. The Lower Cumberland River Watershed is in the Western Pennyroyal Karst Level IV Ecoregion. The geologic base of the watershed is Mississippian-age limestone and includes some chert, shale, siltstone, sandstone, and dolomite. The soils are a thin loess mantle, highly erodible, and very fertile (Baskin et al. 1997). Forests are Western Mesophytic and consist largely of *Quercus* and *Carya* species (Baskin et al. 1997). The watershed encompasses approximately 2,338 square miles and has an estimated population of 155,000 people (TNCT 2015). Over 50% of the watershed is used to produce agriculture products including tobacco, corn, soybean, and livestock (TDEC 2012). The cumulative effects of erosion, agricultural runoff, livestock access to streams, and poorly functioning sewage systems result in poor quality water in the lower reaches of all of the major tributaries in the watershed. The upper reach of Big McAdoo Creek is listed as unimpaired by nutrient enrichment by the Tennessee Department of Environment and Conservation and the United States Environmental Protection Agency, whereas the lower reach is listed as impaired (TDEC 2012). The primary objective of this research was to determine if the upper reach of Big McAdoo Creek should remain listed as unimpaired. We used multiple approaches to evaluate the ecological status of Big McAdoo Creek, including determinations of soluble reactive phosphorous concentration in water samples, evaluations of periphyton biomass and physiological status, and analyses of the structure of the diatom assemblage.

Methods

Periphyton and water were sampled at the lower end of the upper reach of Big McAdoo Creek 100 m downstream of the creek crossing at Hwy 12, 30 km south of Clarksville, Tennessee, on 16 Sep 2015. Two transects from the waterlines of opposing banks, 10 m apart, were established at the sampling site. Transect widths and stream depths at 1/3 intervals of each transect were determined. Stream velocity was determined as the time required for a density-neutral object to travel 10 m downstream. Stream discharge was calculated as: Discharge = Width Depth Velocity 0.9 (Robins & Crawford 1954). A water sample was collected midstream, 5 cm below the surface, to determine the concentration of soluble reactive phosphorous using a Lachat QuickChem 8000 Flow Injection Analyzer (Lachat Instruments, Loveland, Colorado).

Six midstream plots in the 10-m reach were established with 0.25 m^2 wire frames. The fractions of stones designated as very coarse gravel or larger (considered stable substrate) and coarse gravel or smaller (considered unstable substrate) in each plot were recorded. Two cobbles nearest the plot center were removed. One cobble was used for determination of periphyton dry weight, determinations of pigment concentrations, and ash-free periphyton dry weight. One cobble was used to evaluate diatom composition. One sample of unstable substrate from each plot was removed with a core sampler for determinations of pigment concentrations associated with unstable substrate.

Laboratory methods for measurements of ash-free periphyton dry weight and concentrations of periphyton chlorophyll (chl) *a* and pheophytin (pheo) *a* are described in Eaton et al. (2005). The surface area of cobble from which periphyton was removed was calculated by covering the upper surface with aluminum foil, weighing the foil, and extrapolating weight to surface area (Hauer & Lamberti 2006). The autotrophic index (AI) was calculated using the equation of Crossy and LaPoint

(1988): AI = [Ash-free periphyton dry weight (g/m^2)]/[chlorophyll *a* (g/m^2)]. Identification of diatoms and calculation of diatom indices followed the methods described in KDOW (2002) and Lebkuecher et al. (2015).

Results and Discussion

Stream site morphological characteristics were determined to provide detail of the abiotic characteristics of the sampling site (Table 1). Stable substrate for periphyton growth (cobble and very coarse gravel) comprises approximately 1/3 of the benthic environment which is typical for the heterogeneous stream bottoms of mid-order streams in Middle Tennessee (Lebkuecher et al. 2015).

Table 1. Abiotic characteristics of the stream site sampled in Big McAdoo Creek.

Assay	Mean $\pm$ SE
Width (m)	6.5 $\pm$ 0.2
Depth (m)	0.19 $\pm$ 0.02
Velocity (m/s)	1.0 $\pm$ 0.0
Discharge (m^3/s)	0.11 $\pm$ 0.00
Percent benthic substrate	
Stable substrate (cobble + very coarse gravel)	34 $\pm$ 8
Unstable substrate (substrate smaller than very coarse gravel)	66 $\pm$ 9

The concentration of soluble reactive phosphorous of the water (Table 2) was in the range indicative of an oligotrophic to mesotrophic environment (Dodds et al. 1998), however, nutrient concentration of water is often a poor indicator of trophic state. For example, eutrophic environments with excessive concentrations of algae often have very low water phosphorous concentrations due to high phosphorous demand. Periphyton biomass, estimated as the concentration of chl *a* (mg/m^2 stream bottom), at the sampling site (Table 2) is typical of periphyton biomass in other wadeable streams in Tennessee moderately impacted by nonpoint-source pollution (Lebkuecher et al. 2000). The biomass of photoautotrophic periphyton associated with cobble (considered stable substrate) was approximately double that of the biomass associated with course gravel, sand and silt (Table 2). This result supports the conclusion that stable substrate typically supports a greater periphyton biomass relative to unstable substrate (Myers et al. 2007).

Table 2. Concentrations of soluble reactive phosphorous and characteristics of periphyton sampled from Big McAdoo Creek.

Assay	Mean + SE
Soluble reactive phosphorous (mg/L)	0.02
Chl *a* (mg/m^2 stream bottom)	35.5 $\pm$ 2.0
Chl *a* (mg/m^2 cobble)	56.6 $\pm$ 12.2
Chl *a* (mg/m^2 coarse gravel, sand, silt)	28.3 $\pm$ 2.8
Pheo *a* (mg/m^2 cobble)	1.7 $\pm$ 0.8
Ash-free periphyton (g/m^2 cobble)	3.5 $\pm$ 0.6
Autotrophic Index	67 $\pm$ 9

Measurements of the concentration of benthic pheophytin (pheo) *a* reveal the health of photoautotrophic periphyton. Chl *a* is degraded to pheo *a* as photoautotrophic periphyton senesce, hence high concentrations of pheo *a* indicate poor physiological condition. The low concentration of pheo *a* relative to chl *a* (Table 2) indicates the photoautotrophic periphyton were in excellent physiological condition (Eaton et al. 2005). This result suggests the absence of chemical pollutants such as herbicides that adversely affect the health of photoautotrophic periphyton.

Organic pollution results from erosion of organic soil, input of manure or sewage, and overgrowth of algae due to nutrient enrichment (Van Dam et al. 1994). An increase of organic debris can change several biotic characteristics of aquatic environments. Excessive concentrations of decay byproducts such as ammonia and hydrogen sulfide may be harmful to aquatic organisms. The autotrophic index (AI) is the ratio of periphyton biomass (g dry wt/m^2) to photoautotrophic-periphyton biomass (g chl *a*/m^2) and is affected by the concentration of organics (Vannote et al. 1980). AI values typically range from 30 to 300; larger values indicate heterotrophic dominance associated with high concentrations of dissolved organics (Torres-Ruiz et al. 2007). The low AI value of the Big McAdoo site sampled (Table 2) indicates that organic pollution did not impact the structure of the periphyton community.

Forty-two diatom taxa in 17 genera were identified from cobble sampled at the lower end of the upper reach of Big McAdoo Creek (Table 3). The three most abundant diatom taxa were *Achnanthidium rivulare* Potapova and Ponander (28.8 %), *Gomphonema pumilum* (Grun.) Reichardt and Lange-Bert. (18.0 %), and *Achnanthidium minutissimum* (Kütz.) Czarn. (17.8 %). The values for the Shannon Diversity Index and evenness for the diatom assemblage (Table 4) are similar to other diatom assemblages sampled in the ecoregion (Lebkuecher et al. 2013). Despite the high diversity of diatom assemblages in general, dominance by a few taxa is typical and lowers the values for evenness and Shannon Diversity Index.

The Siltation Index (SI) is the percentage of motile diatoms (Bahls 1993). Motile diatoms are able to avoid being buried and are tolerant of sedimentation. SI values $\geq$ 50 denote severely degraded habitat by excessive sediments. The low SI value for the diatom assemblage (Table 4) indicates the reach sampled was not sediment impaired. The organic pollution Index (OPI) is the percentage of taxa tolerant of organic pollution (Kelly 1998). OPI values $\leq$ 20 indicate the absence of significant organic pollution, 21–40 infers some organic pollution present, and values > 40 suggest a significant influence of organic pollution. The low OPI value for the diatom assemblage at the Big McAdoo site (Table 4) indicates the absence of organic pollution. This conclusion is supported by the low value for the Autotrophic Index (Table 2).

The Pollution Tolerance Index of diatom assemblages (PTI) reveals the impact of nutrient concentration on the diatom assemblage and the trophic state of water. The PTI is calculated using the relative abundance and an eutrophication-tolerance value for each taxon of the assemblage. The eutrophication-tolerance value for a taxon is determined from autecological information and ranges from 1 to 4 (KDOW 2002). Taxa very tolerant of eutrophic conditions and more abundant in eutrophic habitats are assigned an eutrophication-tolerance value of 1. Taxa very intolerant of eutrophic conditions are assigned an eutrophication-tolerance value of 4. The PTI ranges from 1 to 4 and values $\leq$ 2.6 indicate a diatom assemblage which is negatively impacted by eutrophication (Lebkuecher et al. 2011). The high PTI value for the diatom assemblage at the Big McAdoo site (2.9; Table 4) indicates the assemblage is not impaired by nutrient enrichment.

Evaluations of the periphyton community at the upper reach of Big McAdoo Creek indicate the site is not markedly impacted by nutrient enrichment. This conclusion is supported by the low concentration of soluble reactive phosphorous of the water, the mesotrophic concentrations of photoautotrophic-periphyton biomass, and the value of the Pollution Tolerance Index of the diatom assemblage. The structure of the diatom assemblage also indicates the assemblage is not impacted by siltation nor dissolved organics. Our results suggest the status of the upper reach of Big McAdoo Creek should remain listed as unimpaired.

Table 3. Diatom taxa and percent taxon composition sampled from cobble in Big McAdoo Creek listed in alphabetical order.

Taxon name	Percent composition
Achnanthidium deflexa Reimer	6.8
Achnanthidium minutissimum (Kütz.) Czarn.	17.8
Achnanthidium rivulare Potapova and Ponander	28.8
Achnanthidium sp.	0.5
Amphora minutissima W. Sm.	0.3
Amphora ovalis (Kütz.) Kütz.	0.8
Amphora perpusilla Grun.	0.3
Amphora veneta Kütz.	0.3
Cocconeis placentula Ehrenb.	0.8
Cocconeis placentula var. euglypta Ehrenb.	0.5
Cymbella affinis Kütz.	2.3
Cymbella tumida (Bréb.) Van Heurck.	1.8
Diploneis eliptica (Kütz.) Cleve	0.3
Encyonema minutum (Hilse) Mann	0.5
Encyonema silesiacum (Bleisch) Mann	0.3
Gomphonema angustatum (Kütz.) Rabenh.	0.8
Gomphonema brasiliense Grun.	1.3
Gomphonema minutum Ag.	1.0
Gomphonema parvulum (Kütz.) Kütz.	0.3
Gomphonema pumilum (Grun.) Reichardt and Lange-Bert.	18.0
Gomphonema sp.	0.8
Gyrosigma acuminatum (Kütz.) Rabenh.	0.3
Karayeva clevei (Grun.) Round and Bukht.	0.3
Melosira varians Ag.	0.3
Navicula capitatoradiata Germ.	0.3
Navicula cryptotenella Lange-Bert.	1.0
Navicula lanceolata (Ag.) Ehrenb.	0.3
Navicula minima Grun.	2.3
Navicula subminuscula Mang.	2.5
Navicula subrotundata Hust.	2.5
Navicula viridula (Kütz.) Ehrenb.	0.5
Nitzchia amphibia Grun.	0.3
Nitzschia dissipata (Kütz.) Grun.	0.3
Nitzschia frustulum (Kütz.) Grun.	0.8
Nitzschia inconspicua Grun.	0.5
Nitzschia linearis (Ag.) W. Sm.	0.3
Nitzschia sinuata var. tabellaria Grun.	1.3
Nitzchia sp.	0.8
Psammothidium curtissimum (Carter) Aboal	0.8
Rhoicosphenia curvata (Kütz.) Grun.	0.3
Sellaphora seminulum (Grun.) D. G. Mann	1.3
Stephanodiscus sp.	0.3

Table 4. Metrics and indices of the diatom assemblage sampled in Big McAdoo Creek. The index scale is the range of values possible from very good to very poor quality water.

Index	Index scale	Index value
Taxa richness		42
Genus richness		17
Shannon Diversity Index		2.5
Evenness	1 - 0	0.7
Siltation Index	0 - 100	13
Organic Pollution Index	0 - 100	12
Pollution Tolerance Index	4 - 1	2.9

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